12 MICROSCOPES

A. All the microscopes used within the Trace Evidence Section are cleaned and serviced annually by qualified service personnel. A log or file of the annual visits is maintained.

B. Lenses are cleaned during use as needed. These include condenser lenses, objective lenses and eyepieces. Miscellaneous microscope maintenance comments are recorded as appropriate.

C. Setting up Köhler illumination is one of the most widely accepted techniques of ensuring that a microscope is correctly aligned, that it has high intensity homogenous illumination and that it is focussed correctly.

D. Depending on a particular microscope and its design, setting up Köhler illumination may be possible in full or only in part. Many modern microscopes are fitted with fixed light sources, which means that it is not possible to center or focus the image of the lamp filament like in conventional microscopes. Also, many modern microscope designs include one or more diffuser filters between the lamp and the condenser. In such cases, even if the microscope is fitted with an adjustable lamp, an image of the filament cannot be seen. In these microscopes only partial Köhler illumination can be obtained.

E. Köhler illumination is a multi-step process, which should be performed or checked every time the microscope is used. Some of the steps should even be checked more often while working.

F. Köhler illumination is set up following the procedures described in the Kodak Publication P-2, *Photography Through the Microscope* by John Delly – 1988 pages 30-35. In short the steps are as follows: (the process is only described for built in illuminators)

a. Center the lamp filament while viewing it with a piece of frosted glass, translucent plastic or paper that is placed on top of the opening in the stand where the light emerges.

b. Focus on a specimen with the different objectives. Start with the lowest power objective.

c. Center all the objectives in the rotating nosepiece individually. (This is only necessary for microscopes with rotating stages.)

d. Adjust the binocular tubes for interpupillary separation and diopter correction.

e. Close the field diaphragm until you see it in the field of view. Focus the image of the field diaphragm by adjusting the substage condenser up or down. Center the focused image by using the centering screws on the condenser. Open the field diaphragm until it is just outside the field of view. (This is the most critical step in setting up Köhler illumination and should be constantly checked while working with the microscope.)

f. Focus the image of the lamp filament, by adjusting the lamp condenser, while viewing it with a Bertrand lens, a pinhole or by removing one eyepiece and looking down the tube. (As mentioned earlier this step may not be possible on some microscopes.) You are now looking at the objective back focal plane.

g. While still viewing the objective back focal plane, close the substage aperture diaphragm until it is visible in the field of view. Center the image of the aperture diaphragm – if possible.

h. Remove the Bertrand lens or pinhole or replace the eyepiece and look at the specimen.

i. Adjust the aperture diaphragm to get the best image contrast. Adjustment of the aperture diaphragm is of utmost importance as it controls the numerical aperture of the system, and therefore the ultimate resolving power, depth of field and contrast of the image. This step should be frequently repeated during work.

♦End